GenCore version 4.5 Copyright (c) 1993 - 2000 Compugen Ltd.

model 3 search, using protein OM protein 3, 2002, 16:27:37; Search time 88.01 Seconds (without alignments) 221.574 Million cell updates/sec Run on:

US-09-534-229C-1 Title:

.....MLGTATGGNLDCYTQRNFAS 1 MARFAALAVCAAALLLAVAA Perfect score:

Scoring table: Sequence:

BLOSUM62 Gapop 10.0 , Gapext 0.5

hits satisfying chosen parameters: ŏ rotal number

219241 seqs, 76174552 residues

Searched:

Minimum DB seq length: 0 Maximum DB seq length: 200000000

Post-processing: Minimum Match 0% Maximum Match 100% Listing first 45 summaries

tinase (EC 3.2.

Chitinase (EC 3.2. 707.5 705.5 705 705 701 701 699 699 

865019

S08627 S56694

T10810

S20982 S20981

S14948

724 723 723 721.5 721.5 711.5

chitinase (EC 3.2.1.14) cht2a precursor - barley C;Species: Hordeum vulgare (barley) C;Date: 10-Dec-1994 #sequence\_revision 10-Nov-1995 #text\_change 22-Jun-1999

R;Bryngelsson, T.; Collinge, D.B.; Green, B.; Gummesson, P.O.; Kragh, K.; Thordal-Cl submitted to the EMBL Data Library, March 1994
A; Description: Purification, characterization and cDNA sequence of a basic chitinas A; Reference number: \$48847

A; Accession: \$48847 A; Molecule type: mRNA A; Mosidues: 1-256 <BRY> A; Cross-references: EMBL: X78671; NID:9563486; PIDN:CAA55344.1; PiD:9563487 C; Genetics:

A;Gene: cht2a A;Gene: cht2a C;Superfamlly: plant chitinase; plant chitinase homology C;Keywords: glycosidase; hydrolase; polysaccharide degradation F;1-26/Domain: signal sequence #status predicted <SIG> F;27-256/Product: chitinase cht2a #status predicted:<AMT> F;31-255/Domain: plant chitinase homology <PCH>

ö Length 256; Query Match 97.9%; Score 1334; DB 2; Length 2 Best Local Similarity 98.4%; Pred. No. 2e-108; Matches 252; Conservative 1; Mismatches 3; Indels

q

61 IAAANTEPGEGTTGSADDIKRDLAAFFGQTSHETTGGTRGAADQFQWGYCFKEEISKATS 120 61 IAAANTFPGFGTTGSADDIKRELAAFFGQTSHETTGGTRGAADQFQWGYCFKEEISKATS 120 δ 셤

121 PPYYGRGPIQLIGRSNYDLAGRAIGKDLVSNPDLVSTDAVVSFRTAMWFWMTAQGNKPSC 180 ò

8

HNVALRRWTPTAADTAAGRVPGYGVITNIINGGLECGMGRNDANVDRIGYYTRYCGMLGT 181 å

181 셤

241 ATGGNLDCYTQRNFAS 256 ò

241

S48848 chitinase (EC 3.2.1.14) cht2b precursor - barley C;Species: Hordeum vulgare (barley)

Class (EC 3.2 (EC 3.2 (EC 3.2 (EC 3.2 (EC 3.2 EC 3.2 ) EC 3.2 (EC 3.2 )

chitinase chitinase chitinase chitinase chitinase chitinase

T10810

65. 64. 63.

11162 11156 11156 11157 11120 1009:5 1007 1001 1001 1004 993

chitinase (EC 3 probable chitin chitinase (EC 3 chitinase (EC 3

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model SV protein search, using protein 3, 2002, 18:32:12 ; Search time 88.01 Seconds (without alignments) 276.101 Million cell updates/sec May

Run on: ĕ

US-09-534-229C-3

1765 1 mrgyvvvamlaaafavsaha......bllgvsygdnidcyngrpfa 319 score: Perfect sc Sequence:

BLOSUM62 Gapop 10.0 , Gapext 0.5 Scoring table:

219241 segs, 76174552 residues ched: 219241 hits satisfying chosen parameters: ö number

seq length: 0 seq length: 2000000000 88 Minimum Maximum

ALIGNMENTS

C;Species: Triticum aestivum (common wheat)
C;Species: Triticum aestivum (common wheat)
C;Date: 20-Feb-1995 #sequence\_revision 20-Feb-1995 #text\_change 22-Jun-1999
C;Accession: S38670
R;Liao, Y.C.; Kreuzaler, F.; Tiburzy, R.; Reisener, H.J.
submitted to the EMBL Data Library, November 1993
A;Reference number: S38670
A;Reference number: S38670 - wheat A; Status: preliminary

A;Molecule type: DNA A;Residues: 1-320 (LinA). A;Cross-references: EMBL:X76041; NID:9416028; PIDN:CAA53626.1; PID:9416029 C;Superfamily: lectin-related plant chitinase; hevein chitin-binding domain homolog: C;Reynords: plycostdase; pdydrolase; polysaccharide degradation E;21-62/Domain: hevein chitin-binding domain homology <HCB> F;82-320/Domain: plant chitinase homology <PCH>

Gaps Length 320; Indels. 96.1%; Score 1696.5; DB 2; 95.3%; Pred. No. 6.8e-119; iive 9; Mismatches 5; 1 Conservative Best Local Similarity Matches 305, Conserv Query Match

1 MRGVVVVAMLAAAFAVSAHAEQCGSQAGGATCPNCLCCSKFGFCGTTSDYCGTGCQSQCN

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7;

9

119 9 GCS-GGTPVPVPTPSGGGVSSIISQSLFDQMLLHRNDAACLAKGFYNYGAFVAAANSFSG 120 FATTGSTDVKKREVAAFLAQTSHETTGGWPTAPDGPYSWGYCFNQERGATSDYCTPSSQW 61 61 g ò a δ

PCAPGKKYFGRGPIQISHNYNYGPAGQAIGTDLLNNPDLVASDATVSFKTALWFWMTPQS 121 180 g ò

PKPSSHDVITGRWSPSGADQAAGRVPGYGVITNIINGGLECGRGQDGRVADRIGFYKRYC 240 ö qq

181

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DLLGVSYGDNLDCYNORPFA 300 301 ö a

RESULT 2 JC2071 chitinase (EC 3.2.1.14) a

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chitinase gene in which the mRNA is extracted from a fully hardened autumn wheat PII73438 (of high snow mould resistance). The genes are useful for creating a plant grade, highly resistant to psychophilic plant pathogenic microbes. 323 AA; Sequence ន្តន្តន្តន្តន្ត

Gaps ö 22; Length 323; Indels Score 1792; DB 22; Pred. No. 2.6e-144; Mismatches 0; ó 100.0%; 100.0%; 323; Conservative Query Match Best Local Similarity Matches

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1 MSTLRARCATAVLAVVLAAAAVTPATAEQCGSQAGGAKCADCLCCSQFGFCGTTSDYCGP 60 

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a ö

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8

PGKQYYGRGPIQLTHNYNYGPAGRAIGVDLLNNPDLVATDPTVAFKTAIWFWMTTQSNKP 240 181 181 셤 ò

SCHDVITGLWTPTARDSAAGRVPGYGVITNVINGGIECGMGQNDKVADRIGFYKRYCDIF 300 셤 ò

GIGYGNNLDCYNQLSFNVGLAAQ 323 301

gigygnnldcynglsfnvglaag 323 301

g

δ

RESULT AAB11489

AAB11489 standard; protein; 319 (first entry) 02-MAR-2001 AAB11489; \* X S X B X B X S X B

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Wheat chitinase protein homologous to spring wheat chitinase.

Wheat; chitinase; low temperature expression; hardened; plant; snow mould resistance; psychophilic plant pathogen; spring wheat.

Triticum aestivum

JP2000270866-A.

03-OCT-2000

99JP-0081694 25-MAR-1999; 99JP-0081694

25-MAR-1999;

(HOKK-) HOKKAIDO NOGKO SHIKENBACHO,

WPI; 2001-027417/04.

New low temperature expression chitinase gene for producing a plant grade highly resistant to psychophilic plant pathogenic microbes

Claim 7; Fig 3; 11pp; Japanese.

This invention describes novel wheat chitinase genes. The invention also describes a method for the isolation of a low temperature expression chitinase gene in which the mRNA is extracted from a fully hardened autumn wheat P1173438 (of high snow mould resistance). The genes are 

New nucleic acid encoding antifreeze polypeptides from plants

Xiong

B,

Moffatt

WPI; 1999-153795/13. N-PSDB; AAX24889. Griffith M, Hew C,

useful for creating a plant grade, highly resistant to psychophilic plant pathogenic microbes. G-----GGGVASIVSRDLFERFLLHRNDAACLARGFYTYDAFLAAAGAFPAFGTTG 122 123 DLDTRKREVAAFFGQTSHETTGGWPTAPDGPFSWGYCFKQEQGSPPSYCDQSADWPCAPG 182 HDVITGLWTPTARDSAAGRVPGYGVITNVINGGIECGMGQNDKVADRIGFYRRYCDIFGI 302 Gaps 64 CHT9; chitinase-like protein; antifreeze protein; AFP; winter rye; antifungal; fungicide; cold tolerance; frost tolerance; transgenic plant; preservation; cryopreservation; tumour; therapy. VLAVVLAAAAVTPATAEQCGSQAGGAKCADCLCCSQFGFCGTTSDYCGPRCQSQCTGCGG 183 KQYYGRGPIQLTHNYNYGPAGRAIGVDLLNNPDLVATDPTVAFKTAIWFWMTTQSNKPSC Length 319 48; Indels 22; 72.2%; Score 1294.5; DB 271.0%; Pred. No. 4.5e-102; 34; Mismatches Rye chitinase-like protein CHT9 preprotein. /note= "signal peptide" Location/Qualifiers AAW98079 standard; Protein; 318 AA. 98WO-CA00745 (first entry) Conservative 303 GYGNNLDCYNQLSF 316 305 sygdnldcyngrpf 318 (ICEB-) ICE BIOTECH INC. CHT9; chitinase-like Similarity Ą; 319 Secale cereale Query Match Best Local Simi Matches 223; WO9906565-A2 31-JUL-1998; 31-JUL-1997; 21-JUN-1999 11-FEB-1999. Sequence AAW98079; Key Peptide Protein 72 243 AAW98079 ខ្លួន្តមូល g ð g 셤 a g ò ò g å ò

particularly with chitinase activity, used to impart frost, and pathogen, resistant to plants, for preservation of foods, cells etc. and for treating tumours

English. Claim 10; Fig 21a; 118pp;

temperatures; to inhibit ice recrystallisation in biological materials or foods; for cryopreservation and hypothermic protection for ells, embryos, tissues etc. (particularly human platelets); and to kill tumour cells. They are also used to inhibit initiation and progression of diseases or spoilage caused by low temperature parbogens (particularly fungi) in plants, frozen foods and any erroperserved biological material. The signal peptide can be used CHT9 preprotein. The mature protein, which is also claimed, is a chitinase-like protein that has chitinase (antifungal) and antifreeze activities. CHT9 CDNA (see AAX24889) was obtained by isolating mRNA from rye plants grown at low temperatures in the absence of pathogens or other stresses, i.e. under conditions when only chitinases with antifreeze activity would be expressed. CHT9 and CHT46 (see AAM98091-83) have been cloned and expressed in bacterial and yeast (Pichia) systems and in Arabidopsis thaliana. The chitinase-like antifreeze proteins can be used: to increase frecing to pleance of plants and microorganisms exposed to sub-zero temperatures; to inhibit ice recrystallisation in biological direct protein secretion in transgenic organisms or expression The signal peptide can be present sequence is winter rye (Secale cereal L.

318 AA; Sequence

Gaps Length 318; Indels 72.0%; Score 1290; DB 20; 71.2%; Pred. No. 1.1e-101; iive 34; Mismatches 48; Conservative Similarity Ma. Local 5. 223; Query Match Best Loca Matches

1;

12 VLAVVLAAAAVTPATAEQCGSQAGGAKCADCLCCSQFGFCGTTSDYCGPRCQSQCTGCGG 71 

tpvpvptptgggvssiisgslfdqmllhrndaaclakgfynygafiaaansfsgfattgg 124 -----GGGGVASIVSRDLFERFLLHRNDAACLARGFYTYDAFLAAAGAFPAFGTTGD 123 72

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LDTRKREVAAFFGQTSHETTGGWPTAPDGPFSWGYCFKQEQGSPPSYCDQSADWPCAPGK 183 125 124 ò

184 QYYGRGPIQLTHNYNYGPAGRAIGVDLLNNPDLVATDPTVAFKTAIWFWMTTQSNKPSCH 243  DVITGLWTPTARDSAAGRVPGYGVITNVINGGIECGMGQNDKVADRIGFYKRYCDIFGIG 303 g ò

304 YGNNLDCYNQLSF 316

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ygdnldcynqrpf 317

AAW98080

AAW98080 standard; Protein; 298 AA

AAW98080,

(first entry)

21-JUN-1999

Rye chitinase-like protein CHT9.

CHT9; chitinase-like protein; antifreeze protein; AFP; winter rye; antifungal; fungicide; cold tolerance; frost tolerance; transgenic plant; preservation; cryopreservation; therapy. 

Xiong F; В, Moffatt 98WO-CA00745 97US-0903872 (ICEB-) ICE BIOTECH INC Hew C, WPI; 1999-153795/13. N-PSDB; AAX24889. Secale cereale. 31-JUL-1997; WO9906565-A2 31-JUL-1998; Griffith M, 

New nucleic acid encoding antifreeze polypeptides from plants -particularly with chitinase activity, used to impart frost, and pathogen, resistant to plants, for preservation of foods, cells etc. and for treating tumours

Claim 10; Fig 21d; 118pp; English

The present sequence is winter rye (Secale cereal L. cv. Muskateer) CHT9 mature protein. It lacks the 20-amino acid signal peptide of the preprotein. (see AAW98079), which is also claimed. Mature CHT9 is a chitinase-like protein that has chitinase (antifungal) and antifreeze activities. CHT9 preprotein CDNA (see AAX24889) was obtained by isolating mRNA from rye plants grown at low temperatures in the absence of pathogens or other stresses, i.e. under conditions when only chitinases with antifreeze activity would be expressed. CHT9 and CHT46 (see AAW98081-82) have been cloned and expressed in bacterial and yeast (Pichla) systems and in Arabidopsis thaliana. The chitinase-ilke antifreeze proteins can be used: to increase freezing tolerance of plants and microorganisms; to increase field survival of plants, animals and microorganisms exposed to sub-zero temperatures; to inhibit ice recrystallisation in biological materials or foods; for cryopreservation and hypothermic protection of cells, embryos, tissues etc. (particularly human platelets); and to kill tumour cells. They are also used to inhibit initiation and progression of diseases or spoilage caused by low temperature pathogens (particularly fungl) in plants, frozen foods and any cryopreserved biological material

298 AA; Sequence

HETTGGWPTAPDGPFSWGYCFKQEQGSPPSYCDQSADWPCAPGKQYYGRGPIQLTHNYNY 199 80 VSRDLFERFLLHRNDAACLARGFYTYDAFLAAAGAFPAFGTTGDLDTRKREVAAFFGQTS 139 Gaps 28 EQCGSQAGGAKCADCLCCSQFGFCGTTSDYCGPRCQSQCTGCGG------GGGGVASI 79 GPAGRAIGVDLLNNPDLVATDPTVAFKTAIWFWMTTQSNKPSCHDVITGLWTPTARDSAA .; ; Length 298; Indels 70.6%; Score 1265; DB 20; 72.7%; Pred. No. 1.3e-99; Mismatches 32; Conservative Query Match Best Local Similarity Matches 216; Conserv 140 121 200

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GRVPGYGVITNVINGGIECGMGQNDKVADRIGFYKRYCDIFGIGYGNNLDCYNQLSF 316 

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